

CLAIMS

1. A biological assay method comprising:
- 5 delivering a sample liquid of a suspension of cells at a controlled steady flow rate through a biochip in the form of an elongate enclosed microchannel;
- 10 causing an externally generated test to be carried out on the sample liquid as it is being delivered through the biochip; and
- examining the sample liquid over time to observe the effect of the test on the sample.
- 15 2. A method as claimed in claim 1 comprising coating the internal bore of the biochip with a protein.
3. A method as claimed in claim 1 comprising coating the internal bore of the biochip with a protein in the form of an extracellular matrix ligand.
- 20 4. A method as claimed in claim 1 comprising:-
- coating the internal bore of the biochip by seeding the biochip with an endothelial cells; and
- 25 allowing the cells to grow and form an endothelial layer on the bore.
5. A method as claimed in claim 1 in which the cell is taken from an animal and the bore of the biochip is substantially the same size as the post capillary venules of the animal.
- 30 6. A method as claimed in claim 1 in which a reagent liquid is delivered simultaneously with the sample liquid through the biochip.

7. A method as claimed in claim 1 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel.
- 5
8. A method as claimed in claim 1, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel, the fluid pressure of the liquids being so chosen as to cause a diffusion of the reagent through the interconnecting channel and into the sample liquid.
- 10
9. A method as claimed in claim 1 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel, and the fluid pressures of the liquids are maintained equal to prevent diffusion of the reagent through the interconnecting channel.
- 15
10. A method as claimed in claim 1 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat.
- 20
11. A method as claimed in claim 1 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat, the fluid pressure of the liquids being so chosen as to cause a diffusion of the reagent through the interconnecting channel and into the sample liquid.
- 25
12. A method as claimed in claim 1 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat and the fluid
- 30

pressures of the liquids are maintained equal to prevent diffusion of the reagent through the interconnecting channel.

13. A method as claimed in claim 1 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat having a cross-section less than that of a cell freely suspended in the sample liquid.
14. A method as claimed in claim 1 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat having a cross-section less than that of a cell freely suspended in the sample liquid, the fluid pressure of the liquids being so chosen as to cause a diffusion of the reagent through the interconnecting channel and into the sample liquid.
15. A method as claimed in claim 1 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat having a cross-section less than that of a cell freely suspended in the sample liquid and the fluid pressures of the liquids are maintained equal to prevent diffusion of the reagent through the interconnecting channel.
16. A method as claimed in claim 1 comprising coating the bore of the microchannel with a hydrophobic coating and delivering a reagent liquid through the microchannel with the sample liquid.
17. A method as claimed in claim 1 comprising coating the bore of the microchannel with a liquid silicone and delivering a reagent liquid through the microchannel with the sample liquid.
18. A method as claimed in claim 1 in which the sample liquid contains more than

one cell type in suspension.

19. A method as claimed in claim 1 in which the sample liquid contains more than one cell type in suspension.

5

20. A method as claimed in claim 1 comprising delivering a reagent liquid and the sample liquid through the microchannel to form multilaminar flow.

21. A method as claimed in claim 1 comprising:-

10

delivering a reagent liquid and a sample through a microchannel to form multilaminar flow, the sample liquid comprising more than one cell type in suspension and the reagent liquid comprising a chemoattractant suitable for one of the types of cell;

15

allowing the flow to continue sufficiently so as to remove that cell type into the reagent liquid; and

separating the reagent liquid and the sample liquid.

20

22. A method as claimed in claim 1 in which the biochip comprises two microchannels, one a feeding microchannel having a cell reservoir intermediate its ends and the other a reactant microchannel connected to the reservoir by a connecting means comprising:-

25

storing cells in the cell reservoir;

feeding and growing the cells in the cell reservoir by delivering a culture medium through the feeding microchannel; and

30

delivering reagent liquid through the reactant microchannel.

23. A method as claimed in claim 1, in which the biochip comprises two microchannels, one a feeding microchannel having a cell reservoir intermediate

its ends and the other a reactant microchannel connected to the reservoir by a connecting means comprising:-

storing cells in the cell reservoir;

5

feeding and growing the cells in the cell reservoir by delivering a culture medium through the feeding microchannel; and

delivering a reagent chosen from one or more of a chemoattractant toxic substance and pharmaceutical preparation recombinant or cell derived through the reactant microchannel.

10

24. A method as claimed in claim 1 in which a plurality of tests are carried out simultaneously using a sample liquid forming portion of a larger sample and using different test conditions.

15

25. A method as claimed in claim 1 in which a plurality of tests are carried out simultaneously using different sample liquids and the same test conditions.

20

26. A biological assay method one or more of cell flow; rolling; binding; tethering and migration of previously adhered cells and adhesion comprising:-

preparing a sample liquid of a suspension of animal cells;

25

coating the internal bore of a biochip with a protein, the biochip comprising an elongate enclosed microchannel having a bore substantially the same size as the post capillary venules of the animal;

delivering the sample liquid at a controlled steady flow rate through the microchannel; and

30

observing the effect of the test over time.

27. A biological assay method as claimed in claim 26, comprising coating the

internal bore of the biochip with a protein in the form of an extracellular matrix ligand.

28. A biological assay method as claimed in claim 26, comprising:-

coating the internal bore of the biochip by seeding the biochip with an endothelial cells; and

allowing the cells to grow and form an endothelial layer on the bore.

29. A biological assay method as claimed in claim 26, in which the cell is taken from an animal and the bore of the biochip is substantially the same size as the post capillary venules of the animal.

30. A biological assay method as claimed in claim 26, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel.

31. A biological assay method as claimed in claim 26, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel, the fluid pressure of the liquids being so chosen as to cause a diffusion of the reagent through the interconnecting channel and into the sample liquid.

32. A biological assay method as claimed in claim 26, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel, and the fluid pressures of the liquids are maintained equal to prevent diffusion of the reagent through the interconnecting channel.

33. A biological assay method as claimed in claim 26, delivering a reagent liquid at

a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat.

- 5 34. A biological assay method as claimed in claim 26, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat, the fluid pressure of the liquids being so chosen as to cause a diffusion of the reagent through the interconnecting channel and into the sample liquid.
- 10
35. A biological assay method as claimed in claim 26, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat and the fluid pressures of the liquids are maintained equal to prevent diffusion of the reagent through the interconnecting channel.
- 15
36. A biological assay method as claimed in claim 26, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat having a cross-section less than that of a cell freely suspended in the sample liquid.
- 20
37. A biological assay method as claimed in claim 26, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat having a cross-section less than that of a cell freely suspended in the sample liquid, the fluid pressure of the liquids being so chosen as to cause a diffusion of the reagent through the interconnecting channel and into the sample liquid.
- 25
- 30

38. A biological assay method as claimed in claim 26, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat having a cross-section less than that of a cell freely suspended in the sample liquid and the fluid pressures of the liquids are maintained equal to prevent diffusion of the reagent through the interconnecting channel.
39. A biological assay method as claimed in claim 26, comprising coating the bore of the microchannel with a hydrophobic coating and delivering a reagent liquid through the microchannel with the sample liquid.
40. A biological assay method as claimed in claim 26, comprising coating the bore of the microchannel with a liquid silicone and delivering a reagent liquid through the microchannel with the sample liquid.
41. A biological assay method as claimed in claim 26, in which the sample liquid contains more than one cell type in suspension.
42. A biological assay method as claimed in claim 26 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat having a cross-section less than that of a cell freely suspended in the sample liquid.
43. A biological assay method as claimed in claim 26 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat having a cross-section less than that of a cell freely suspended in the sample liquid, the fluid pressure of the liquids being so chosen as to cause a diffusion of the reagent through the interconnecting channel and into the sample liquid.



- 5 44. A biological assay method as claimed in claim 26 comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel having a restricted entry throat having a cross-section less than that of a cell freely suspended in the sample liquid and the fluid pressures of the liquids are maintained equal to prevent diffusion of the reagent through the interconnecting channel.
- 10 45. A biological assay method as claimed in claim 26 comprising coating the bore of the microchannel with a hydrophobic coating and delivering a reagent liquid through the microchannel with the sample liquid.
- 15 46. A biological assay method as claimed in claim 26 comprising coating the bore of the microchannel with a liquid silicone and delivering a reagent liquid through the microchannel with the sample liquid.
- 20 47. A biological assay method as claimed in claim 26, in which the sample liquid contains more than one cell type in suspension.
48. A biological assay method as claimed in claim 26, comprising delivering a reagent liquid and the sample liquid through the microchannel to form multilaminar flow.
- 25 49. A biological assay method as claimed in claim 26, comprising:-
- 30 delivering a reagent liquid and a sample through a microchannel to form multilaminar flow, the sample liquid comprising more than one cell type in suspension and the reagent liquid comprising a chemoattractant suitable for one of the types of cell;
- allowing the flow to continue sufficiently so as to remove that cell type into the reagent liquid; and

separating the reagent liquid and the sample liquid.

50. A biological assay method as claimed in claim 26, in which the biochip comprises two microchannels, one a feeding microchannel having a cell reservoir intermediate its ends and the other a reactant microchannel connected to the reservoir by a connecting means comprising:-

storing cells in the cell reservoir;

- 10 feeding and growing the cells in the cell reservoir by delivering a culture medium through the feeding microchannel; and

delivering reagent liquid through the reactant microchannel.

- 15 51. A biological assay method as claimed in claim 26, the biochip comprises two microchannels, one a feeding microchannel having a cell reservoir intermediate its ends and the other a reactant microchannel connected to the reservoir by a connecting means comprising:-

- 20 storing cells in the cell reservoir;

feeding and growing the cells in the cell reservoir by delivering a culture medium through the feeding microchannel; and

- 25 delivering a reagent chosen from one or more of a chemoattractant toxic substance and pharmaceutical preparation recombinant or cell derived through the reactant microchannel.

52. A biological assay method as claimed in claim 26, in which a plurality of tests are carried out simultaneously using a sample liquid forming portion of a larger sample and using different test conditions.

53. A biological assay method as claimed in claim 26, in which a plurality of tests are carried out simultaneously using different sample liquids and the same test

conditions.

54. A transmigration assay to determine cell migration from the endothelium to the extracellular matrix comprising:-

5

delivering a sample liquid comprising a suspension of cells at a controlled steady flow rate through a microchannel forming part of a biochip;

10

delivering a chemoattractant through another microchannel forming part of the biochip and being connected to the other microchannel through a restricted entry of size less than that of a freely suspended cell; and

15

observing the migration of cells through the restricted entry to the chemoattractant.

55. An assay as claimed in claim 54, comprising coating the internal bore of the biochip with a protein.

20

56. An assay as claimed in claim 54, comprising coating the internal bore of the biochip with a protein in the form of an extracellular matrix ligand.

57. An assay as claimed in claim 54, comprising:-

25

coating the internal bore of the biochip by seeding the biochip with an endothelial cells; and

allowing the cells to grow and form an endothelial layer on the bore.

30

58. A method as claimed in claim 54, in which the fluid pressure of the liquids is so chosen as to provide a diffusion of the sample liquid through the restricted entry.

59. A method as claimed in claim 54 in which the fluid pressure of the liquids is the same.
- 5 60. A method as claimed in claim 54, in which a plurality of tests are carried out simultaneously using a sample liquid forming portion of a larger sample and using different test conditions.
61. A method as claimed in claim 54, in which a plurality of tests are carried out simultaneously using different sample liquids and the same test conditions.
- 10 62. A method of separating one cell type from a sample liquid containing more than and at least one other one cell type comprising:-
- 15 delivering a chemoattractant and the sample liquid through a microchannel forming part of a biochip, the liquids forming multilaminar flow and the chemoattractant having an affinity to the cell type allowing the flow to continue sufficiently so as to remove that cell type into the chemoattractant; and
- 20 subsequently separating the chemoattractant liquid and the sample liquid.
63. A method as claimed in claim 62, in which a plurality of tests are carried out simultaneously using a sample liquid forming portion of a larger sample and using different test conditions.
- 25 64. A method as claimed in claim 62, in which a plurality of tests are carried out simultaneously using different sample liquids and the same test conditions.
- 30 65. A biochip comprising:-
- an elongate main microchannel;
- an inlet port mounted on the proximal end of the main microchannel;

an outlet port adjacent its distal end;

a separate liquid feeder microchannel connected to the main microchannel, the feeder microchannel having an inlet port; and

an outlet feeder port connecting the feeder microchannel and the main microchannel.

66. A biochip as claimed in claim 65 in which the outlet port between the feeder microchannel and the main microchannel has a restricted throat.
67. A biochip as claimed in claim 65, in which a microchannel connects to a further take-off microchannel intermediate its ends.
68. A biochip as claimed in claim 65, in which the microchannel comprises a main microchannel and take-off microchannel intermediate its ends, the take-off microchannel having an entrance which projects into the main microchannel to divert flow from the main microchannel into the take-off microchannel.
69. A biochip as claimed in claim 65 comprising a microwell connected to a microchannel forming part of the biochip.
70. A biochip as claimed in claim 65 in which there is a microwell connected to the microchannel feeder delivering into and out of the microwell, the feeder microchannel having an inlet port adjacent its proximal end and an outlet port adjacent its distal end.
71. A biochip as claimed in claim 65 in which the microchannel comprises a planar top wall.
72. A biochip as claimed in claim 65 in which the microchannel comprises planar top, bottom and side walls.

73. A biochip as claimed in claim 65 in which the side walls taper outwardly and upwardly away from each other.
74. A biochip as claimed in claim 65 in which the top wall is removable.
- 5 75. A biochip as claimed in claim 65 in which the microchannel side and bottom walls are formed in a planar sheet of plastics material and the top wall is formed by a plastics film adhered to the sheet.
- 10 76. A biochip as claimed in claim 65 in which each port has a bubble release port and a valve associated therewith.
77. A biochip as claimed in claim 65 in which the cross-sectional area of the microchannel is between  $25 \mu\text{m}^2$  to  $10,000 \mu\text{m}^2$ .
- 15 78. A biochip as claimed in claim 65 in which the cross sectional area of the microchannel is in excess of  $400 \mu\text{m}^2$ .
79. A biochip assembly comprising a plurality of biochips as claimed in claim 65 formed on the one base sheet.
- 20 80. A biochip assembly as claimed in 65 comprising a plurality of biochips formed on the one base sheet with a common feeder microchannel having a port therein connected to each of the biochips.
- 25 81. A biochip comprising:-
- two separate elongate main microchannels,
- 30 a connecting microchannel between the two separate main microchannels,
- an inlet port mounted on the proximal end of each of the main microchannels, and

an outlet port mounted on the distal end of each microchannel.

5 82. A biochip as claimed in claim 81 comprising a separate liquid feeder microchannel connected to at least one main microchannel, the feeder microchannel having an inlet port and an outlet feeder port connecting the feeder microchannel and the main microchannel.

10 83. A biochip as claimed in claim 81, comprising:-  
coating the internal bore of the biochip by seeding the biochip with an endothelial cells; and

15 allowing the cells to grow and form an endothelial layer on the bore.

84. A biochip as claimed in claim 81 in which the cell is taken from an animal and the bore of the biochip is substantially the same size as the post capillary venules of the animal.

20 85. A biochip as claimed in claim 81, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel.

25 86. A biochip as claimed in claim 81, comprising delivering a reagent liquid at a controlled steady flow rate through another microchannel connected to the other microchannel, the channels being connected intermediate their ends by an interconnecting channel, the fluid pressure of the liquids being so chosen as to cause a diffusion of the reagent through the interconnecting channel and  
30 into the sample liquid.

87. A biochip as claimed in claim 81, in which the sample liquid contains more than one cell type in suspension.

88. A biochip as claimed in claim 81, in which the top wall is removable.
89. A biochip as claimed in claim 81, in which the microchannel side and bottom walls are formed in a planar sheet of plastics material and the top wall is formed by a plastics film adhered to the sheet.
90. A biochip as claimed in claim 81, in which each port has a bubble release port and a valve associated therewith.
91. A biochip as claimed in claim 81, in which the cross-sectional area of the microchannel is between  $25\text{ }\mu\text{m}^2$  to  $10,000\text{ }\mu\text{m}^2$ .
92. A biochip assembly comprising a plurality of biochips as claimed in claim 81 formed on the one base sheet.
93. A biochip assembly as claimed in claim 81, comprising a plurality of biochips formed on the one base sheet with a common feeder microchannel having a port therein connected to each of the biochips.
94. A biochip comprising:-
- two separate elongate main microchannels;
  - a common microchannel connected to each main channel adjacent their distal and its proximal end; and
  - an inlet port mounted on the proximal end of each of the main microchannels.
95. A biochip as claimed in claim 94, comprising a separate liquid feeder microchannel connected to at least one main microchannel, the feeder microchannel having an inlet port and an outlet feeder port connecting the feeder microchannel and the main microchannel.



96. A biochip as claimed in claim 94, in which the common microchannel feeds two further main microchannels.

5 97. A biochip as claimed in claim 94, in which a microchannel connects to a further take-off microchannel intermediate its ends.

10 98. A biochip as claimed in claim 94, in which the microchannel comprises a main microchannel and a take-off microchannel intermediate its ends, the take-off microchannel having an entrance which projects into the main microchannel to divert flow from the main microchannel into the take-off microchannel.

99. A biochip as claimed in claim 94, in which the microchannel comprises planar top, bottom and side walls.

15 100. A biochip as claimed in claim 94, in which the microchannel side and bottom walls are formed in a planar sheet of plastics material and the top wall is formed by a plastics film adhered to the sheet.

20 101. A biochip assembly comprising a plurality of biochips as claimed in claim 94 formed on the one base sheet.

25 102. A biochip assembly as claimed in claim 94 comprising a plurality of biochips formed on the one base sheet with a common feeder microchannel having a port therein connected to each of the biochips.

103. A biochip comprising:-

an elongate main microchannel;

30 an inlet port mounted on the proximal end of the main microchannel;

an outlet port adjacent its distal end;

a microwell connected to the main microchannel;

a feeder microchannel for delivering liquid into and out of the microwell;

an inlet port adjacent the proximal end of the feeder microchannel; and

an outlet port adjacent the distal end of the feeder microchannel.

104. A biochip as claimed in claim 103, in which the microchannel comprises planar top, bottom and side walls.

105. A biochip as claimed in claim 103, in which the microchannel side and bottom walls are formed in a planar sheet of plastics material and the top wall is formed by a plastics film adhered to the sheet.

106. A biochip as claimed in claim 103, in which each port has a bubble release port and a valve associated therewith.

107. A biochip assembly comprising a plurality of biochips as claimed in claim 103 formed on the one base sheet.

108. A biochip assembly as claimed in claim 103 comprising a plurality of biochips formed on the one base sheet with a common feeder microchannel having a port therein connected to each of the biochips.